agriculture

MDPI

*Article*

# Linking Sap Flow and Trunk Diameter Measurements to Assess Water Dynamics of Touriga-Nacional Grapevines Trained in Cordon and Guyot Systems

**Aureliano C. Malheiro [1,*], Mafalda Pires [1], Nuno Conceição [2], Ana M. Claro [1], Lia-Tânia Dinis [1] and José Moutinho-Pereira [1]**

1 Centre for the Research and Technology of Agro-Environmental and Biological Sciences (CITAB), University of Trás-os-Montes e Alto Douro, 5000-801 Vila Real, Portugal; mafaldap@utad.pt (M.P.); anaclaro@utad.pt (A.M.C.); liatdinis@utad.pt (L.-T.D.); moutinho@utad.pt (J.M.-P.)

2 Linking Landscape, Environment, Agriculture and Food (LEAF), University of Lisbon, 1349-017 Lisbon, Portugal; nuconceicao@isa.ulisboa.pt

* Correspondence: amalheir@utad.pt

Received: 30 June 2020; Accepted: 26 July 2020; Published: 1 August 2020

check for updates

**Abstract:** The present research aimed to evaluate the water dynamics of grapevines trained in Cordon and Guyot systems by coupling sap flow and trunk diameter measurements under Mediterranean climate conditions. The study was conducted in a vineyard with Touriga-Nacional located at the Douro Valley, Portugal, during 2017. The results showed daily trunk diameter fluctuations (TDFs), with the contraction, recovery and increment phases and higher sap flow (SF) rates at earlier stages. Under harsh pedoclimatic conditions, SF was reduced and TDF flattened. Rehydration and stomatal mechanisms were mostly associated with these responses. Guyot vines showed higher changes in TDF for the same SF values, whereas the TDFs of Cordon vines remained practically unchanged over maturation. Guyot vines generally showed increased values of cumulative increment and maximum daily trunk shrinkage. Although Guyot vines had a similar leaf area index (LAI), they showed higher SF/LAI ratios than Cordon vines. These results highlight the effect of the shorter length of the hydraulic pathways of the Guyot training system, in contrast to the higher trunk and the permanent horizontal branch (cordon) of the Cordon training system, indicating good adaptation to local pedoclimatic conditions. The study pointed to the complementary use of both techniques in the evaluation of grapevine water dynamics.

**Keywords:** Douro Demarcated Region; plant-based sensors; training system; *Vitis vinifera*

## 1. Introduction

*Vitis vinifera* is an environmentally sensitive species, mainly to climate and soil conditions [1]. Radiative, thermal and water stresses, generally combined, are the main causes of reduced growth and yield of vineyards in Mediterranean winegrowing regions, particularly under rain-fed conditions [2,3]. Furthermore, these regions, characterized by long growing seasons, mild-warm temperatures and dry summers [4,5] are likely to face a greater and intensified number of abiotic events in the near future [6,7].

Depending on the crop and variety, plants can control their water status by developing strategies to cope with critical conditions of atmospheric evaporative demand and soil water availability [8,9]. Water redistribution within the plant has been identified as a crucial survival strategy [10,11]. However, the mechanism generally considered most important for plant survival in a hot and dry environment is stomatal control to reduce transpiration [3,4,12,13].

Adaptation strategies involving cultural practices must be adopted to guarantee economic, social and environmental sustainability [2,7]. One major adaptation strategy is the selection of the grapevine training system [14,15]. The proper training system option helps to control and regulate canopy growth, improve leaf area exposure to maximize light interception, form a renewal zone to ensure that the shape and yield of the vine are maintained, improve potential production and facilitate mechanization and disease control [16,17]. The different training systems cause changes in the microclimate [14,18] that can also influence bud differentiation, bunch exposure, grapevine water status and leaf transpiration [16]. Some studies have reported that grapevine training systems have significant effects on the berry quality, such as pH, sugar content, anthocyanins and phenols [19].

Among the Portuguese land areas devoted to wine production, the Douro valley (NE Portugal) stands out as being the first in the world to be demarcated and regulated as a Wine Region (1756), producing the world-famous Port wine and other high-quality table wines. Most importantly, the Douro Demarcated Region (DDR) ranks first place in vineyard area, volume production and wine export value in national terms [20]. The Cordon is currently the most widely used training system in the DDR [21]. This training system (spur pruning in vertical shoot positioned) is relatively easy to define, simple to prune in winter and is adapted to mechanization [16]. However, a training system with a lower trunk, such as the Guyot system (cane pruning in vertical shoot positioned, which was traditionally used in the DDR before the 1980s), may bring advantages in hot and dry areas such as the DDR [21]. In addition, this author highlights how plants trained in the Guyot system have less overlapping of pruning cuts, leading to lower susceptibility to trunk diseases and greater longevity.

The use of plant-based measurement sensors in agriculture has increased as a result of technological advances and with a greater focus on spatial management of crop inputs [9]. The use of relatively simple, reliable and non-destructive methods of continuous monitoring of the plant water dynamics allows us to evaluate the influence of various environmental factors and the subsequent crop performance [22,23]. Linear variable displacement transducer sensors (LVDTs) permit continuous measurement of trunk diameter fluctuations (TDFs), which may be related to plant growth, use and water status [9,12,24]. TDFs act in response to the water content in trunk tissues, caused by the reversible contraction and expansion of dead and living tissues and irreversible radial growth [25]. The daily TDF cycle can provide indices, such as trunk growth rate and maximum daily trunk shrinkage [26–28], frequently computed for irrigation scheduling. Sap flow (SF) measurements reflect the flow of water from the soil to the plant (within the xylem tissue) and into the atmosphere through the stomata (whole-plant transpiration) without disturbing the leaf environment [11,12,24], provided that adequate corrections are performed [5]. SF determinations can be completed by methods such as the thermal dissipation technique [29], which stands out for its simplicity and relatively low cost [5].

The coupling of SF measurements with quantification of TDF has been reported for various crops (e.g., in tomato plants [30], lemon trees [31,32], peach trees [33,34] and olive trees [35,36]. For grapevines, different studies were carried out separately using SF (e.g., Ferreira et al. [5]) and TDF measurements (e.g., Intrigliolo and Castel [37]). The few studies that combined both techniques were developed in potted grapevines [24] and in field work were focused on statistical methods for stress detection [38] and on the relationship with the reference evapotranspiration [39].

Therefore, the objectives of the present study were threefold: (i) to evaluate the daily and seasonal water dynamics of Touriga-Nacional grapevines trained in the Guyot and Cordon systems throughout the growing season, (ii) to assess their responses to water supply events and (iii) to appraise the joint use of sap flow and trunk diameter measurements. Touriga-Nacional is considered Portugal's "finest" and most international red variety, ranking in third place in terms of national surface area (following Aragonez and Touriga-Franca), and is extensively planted in the DDR (about 10%), among more than 100 authorized (mostly native) varieties [20,21].

## 2. Material and Methods

### 2.1. Site Description and Plant Material

The study was carried out in a commercial vineyard, located at the Upper Douro sub-region (the eastern sub-region with harsher climate conditions) of the DDR, Portugal (41°04′18″ N, 7°04′51″ W, 160 m) during the 2017 growing season. The climate of this area is typically Mediterranean (warm temperate climate with dry and hot summer; see [40]) with a long-term average annual rainfall of 580 mm year$^{-1}$ (mostly concentrated in the winter months) and average annual minimum and maximum temperatures of 10.0 and 19.8 °C respectively [41]. Meteorological variables were monitored by an automatic weather station (IMT280, iMETOS, Pessl Intruments, Weiz, Austria) positioned at the experimental site. From the weather data, reference evapotranspiration ($ET_0$) following the FAO Penman–Monteith method [42] and vapor pressure deficit (VPD) were calculated.

*Vitis vinifera* L. Touriga-Nacional grafted onto 110 Richter rootstocks were planted in 2011 at a vertical (30% slope) spacing of 2.2 × 1.0 m (4545 vines ha$^{-1}$), with E-W orientation. Two training systems were defined: unilateral (single) Cordon (trunk height of about 0.6 m) and single Guyot (trunk height of about 0.4 m) systems with similar numbers of buds (eight) left at winter pruning. From the selected field area, the closest six training consecutive rows (with just one path between training systems) were chosen, of which only the two center rows were used for measurements. The soil, essentially of schist origin with a loam-dominated texture, is classified as surribi-aric anthrosol [43].

The vineyard was managed (similarly in both training systems, e.g., soil fertilization) according to the grower's commercial cultural practices. Between rows, a cover crop was mowed regularly that became senescent during summer. To prevent plant death, around 6-mm events of water (from end-June to late-August; Figure 1), totaling 50 mm, were provided. For this purpose, a drip irrigation system was positioned in the center of the row, consisting of 2.2 L h$^{-1}$ self-compensating drippers, spaced 1.0 m apart.

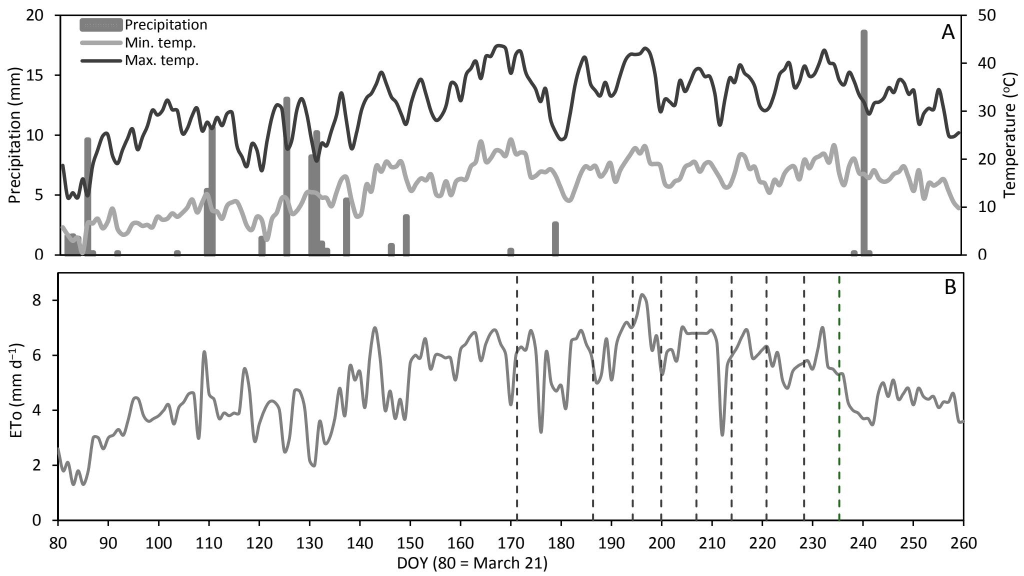


**Figure 1.** Precipitation and maximum and minimum air temperatures at the experimental site in 2017 (**A**). Daily values of reference evapotranspiration ($ET_0$) were computed from records of the weather station. The dashed lines indicate an irrigation event (**B**).

### 2.2. Sap Flow Measurements

The thermal dissipation technique [29,44] was used to continuously monitor SF within the xylem of Cordon and Guyot-trained vines over the growing season. Five vines per training system were chosen and the sensors installed in the north facing side of the grapevine's trunk. The SF system consisted of a pair of cylindrical probes, one heated and the other unheated, and an electrical current supplier (UP GmbH, Ibbenbüren, Germany). The probes were installed in smooth internode tissue of the trunk, avoiding irregular tissue at the nodes. The trunk was thermally insulated with standard styrofoam pipe, extending approximately 0.15 m above and below the probes. Reflecting material (thick aluminum foil) was applied around the probes and trunk (down to the ground) to thermally insulate the system and to minimize the formation of natural thermal gradients. In addition, four non-heated sensor-pairs per training system were installed in other selected vines (avoiding thermal effects between different sensors) in order to account for the remaining natural gradients. In this way, unheated patterns were applied to the heated sets and corrected temperature differences were then used [5]. A data logger (CR1000 with an AM25T multiplexer, Campbell Scientific Inc., Shepshed, UK) was used for measuring the temperature differential between thermocouples every 30 s, programmed to store mean values every 5 min.

### *2.3. Trunk Diameter Measurements*

Trunk diameter fluctuations (TDF) were measured using linear variable differential transformers (LVDT; Solartron Metrology model DF ± 2.5 mm, accuracy ± 10 μm, Bognor Regis, UK). Six sensors were installed, divided by both training systems, which had been previously calibrated. Each sampling grapevine was equipped with a sensor, fixed to the main trunk with a bracket made of aluminum, with a thermal expansion close to zero [45,46]. The sensors were covered with thermo-protected silver sheets to prevent the device from heating up and wetting. Readings were taken every 30 s with a CR1000 Campbell datalogger combined with an AM16/32 Campbell multiplexer, programmed to report values every 5 min. Indices from the TDF daily cycles were derived, such as maximum (MXTD) and minimum (MNTD) daily trunk diameters, cumulative increment (CI) and maximum daily trunk shrinkage (MDS). The CI was calculated as the cumulated difference between MXTD of a given day and MXTD of the previous day and MDS as the difference between MXTD and MNTD of the defined day [47].

### *2.4. Other Measurements*

Soil water content was periodically measured within a row of each training system (with five replicates). For this purpose, 0.6-m buriable waveguides were installed and periodically connected to a portable TDR (miniTrase, Soilmoisture Equipment Corp., Santa Barbara, CA, USA). At a given date, the fraction of available water (FASW) within the defined soil profile (0–0.6 m) was calculated from the soil water content on the day of measurement and the soil water content at field capacity and minimum soil water content in situ [4].

Determinations of predawn leaf water potential ($\Psi_{pd}$) were performed on cloud free days, between mid-June and late-August (harvest), at 5:30 (completed before sunrise). For this, six fully expanded leaves were collected from the outside of the south-facing middle third of the canopy of distinct vines. Immediately, they were introduced in a Scholander Pressure Chamber (model 1505D-EXP, PMS Instrument, Albany, OR, USA). Measurements were performed according to Scholander et al. [48] and following the recommendations of Turner [49].

The leaf area index (LAI) was computed from measuring transmitted photosynthetically active radiation along all the inter-row space (readings were taken perpendicular to rows) by a ceptometer (AccuPAR model LP-80, Pullman, WA, USA), as described by López-Lozano and Casterad [50]. From early-July to late-August, the measuring process was replicated 10 times in each training system, with a 0.5-m row spacing between readings. Estimated LAI values were then averaged to calculate the integrated canopy LAI.

During the irrigation periods, all these measurements were taken 1–2 days before the water supply.

### *2.5. Statistical Analysis*

Statistical analysis of FASW, $\Psi_{pd}$, LAI and SF/LAI data was performed using the SPSS software, version 24.0 (IBM Corporation, Armonk, NY, USA). Analysis of variance (ANOVA) was performed for each of the dependent variables studied, with a confidence interval of 95%, followed by the Tukey test to compare significantly different means.

## 3. Results

### *3.1. Weather Conditions*

From budburst to harvest (late-March to end of August), daily maximum temperature ranged from 12.0 °C (DOY 81) to 43.6 °C (DOY 168) and daily minimum temperature varied between 0.5 °C (DOY 84) and 24.1 °C (DOY 170). There were several days, from June to August, with mean temperatures above 30 °C (Figure 1A, Figure S1). Conversely, precipitation was very low or even zero during these months. Nevertheless, there was an atypical rainy day on August 29 (19 mm) at harvest time. The total annual (November 2016–October 2017, Figure S1) precipitation was 320 mm, of which 25% was observed during the growing season (corresponding to about 70% from November 2016 to March 2017). The reference evapotranspiration ($ET_0$) varied between 1.3 (DOY 83) and 8.2 mm $d^{-1}$ (DOY 196) from end-March to late-August, with a total amount of 805 mm for this time period (Figure 1B) and an annual sum of 1140 mm.

### *3.2. Fraction of Available Soil Water and Predawn Leaf Water Potential*

The local weather conditions had a clear impact on the fraction of available soil water, FASW (Figure 2A). In fact, FASW sharply declined from 55% (mid-May) to 30% in about two weeks. Despite the water provided by irrigation and precipitation (e.g., DOY 179) that caused occasional increases in FASW, this variable globally diminished to a minimum of about 20% in August (maturation). No significant FASW differences were found between training systems. Similarly, predawn leaf water potential ($\Psi_{pd}$) data revealed no significant differences between training systems (Figure 2B). The $\Psi_{pd}$ generally decreased from −0.40 MPa (mid-June) to around −0.85 MPa at the beginning of August (maturation), followed by a late recovery at harvest ($\Psi_{pd}$ = −0.35 MPa) due to a rainfall event. However, there were no substantial leaf losses (visual observation) owing to sunburn over the growing cycle.

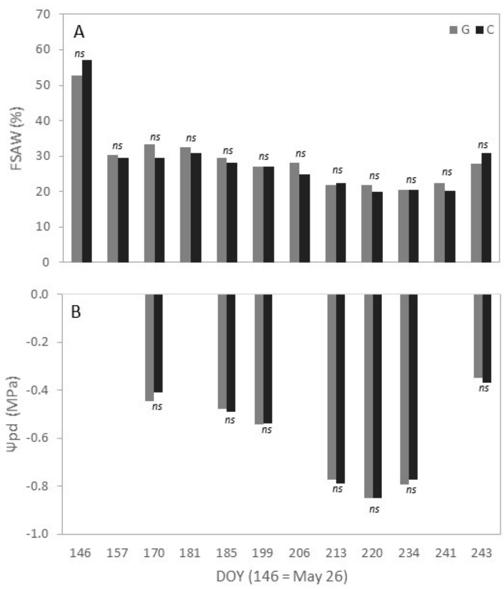


**Figure 2.** Fraction of available soil water (FASW) (**A**) and predawn leaf water potential ($\Psi_{pd}$) (**B**) measured in Touriga-Nacional grapevines trained in Cordon (C) and Guyot (G) systems. *ns*, not significant.

### *3.3. Sap Flow and Trunk Diameter Fluctuations*

In terms of plant water dynamics, Figure 3A–F show the daily courses of SF and TDF values during three phenological stages: post-flowering (Figure 3A,D), post-veraison (Figure 3B,E) and mid-maturation to ripening (Figure 3C,F). Generally, both variables exhibited higher values at the earlier phase, decreasing in the following, being clearer in the TDF values (Figures 3 and S2). These patterns were consistent with FASW and $\Psi_{pd}$ trends.

During the first interval (Figure 3A,D), daily SF displayed a typical bell-shaped curve that increased markedly from sunrise until noon (generally following solar radiation with maximum values reaching 1000 $Wm^{-2}$, Figure 3G), falling to negligible values at the end of the day. These SF maximums became lower over the maturation period (Figure 3B,C,E,F). Although irrigation tended to increase SF, the maximum values were lower in the later periods than in the first interval. This reduction in SF rates was a reflection of the progressive soil water depletion (decreasing FASW) and enhanced atmospheric demand (Figure 3H,I), as well as the decrease in total leaf area by the onset of senescence towards the harvest period (as shown in Section 3.4). Daily maximum values of vapor pressure deficit (VPD) and reference evapotranspiration ($ET_0$) peaked at 7.6 kPa and around 8.0 mm, respectively, during maturation (Figure 3H–I).

TDF data showed a clear daily cycle, with increasing values until the early morning, followed by a decreasing period and, lastly, a recovery (Figure 3A–F). However, TDFs were higher and displayed a positive trend at the early stage (Figure 3A,D), indicating trunk increment during these days. This trend did not occur, and even became negative (no trunk increment), later on (Figure 3B,C,E,F). This negative trend was moderately sustained and occasionally reversed due to the water supply by irrigation.

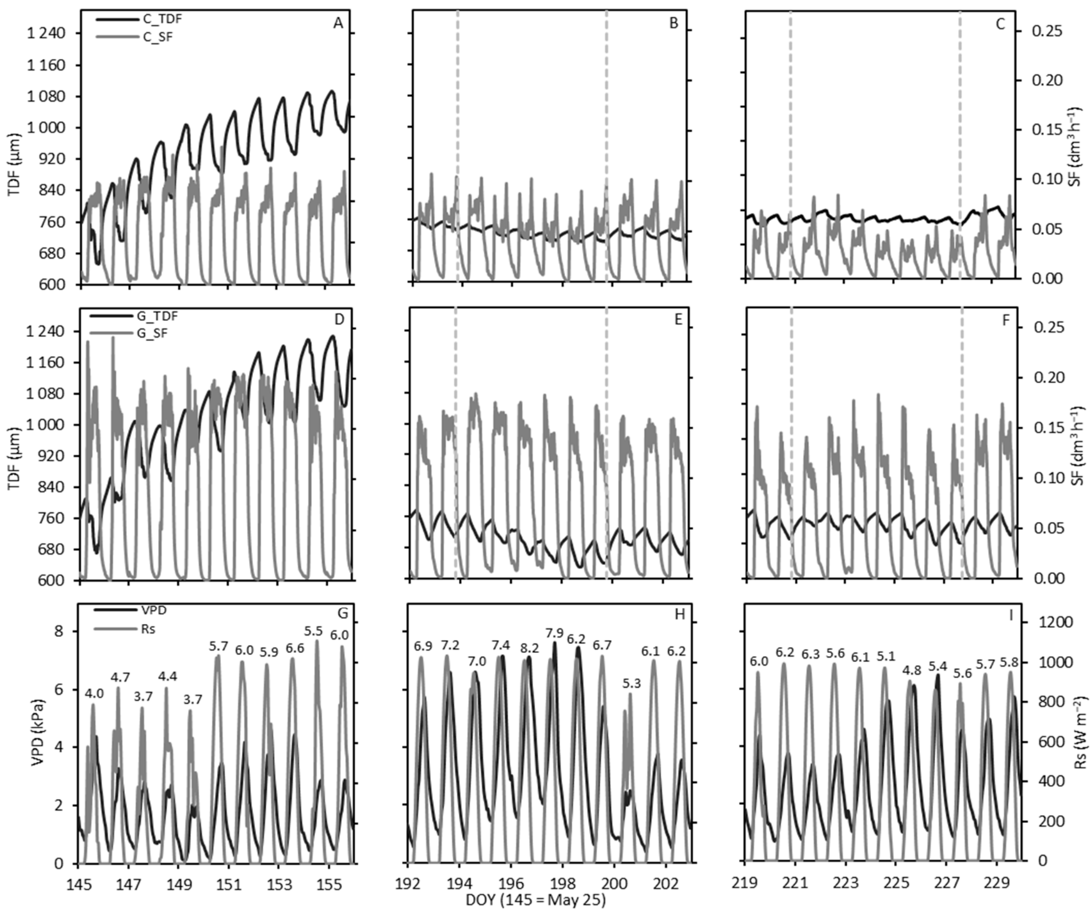


**Figure 3.** Time courses of sap flow (SF) and trunk diameter fluctuation (TDF) in Touriga-Nacional trained in Cordon (C) system for post-flowering (**A**), post-veraison (**B**) and mid-maturation to ripening stages (**C**). Similarly, for Guyot (G) trained vines (**D**–**F**). The dashed lines (in B, C, E and F) indicate an irrigation event. (**G**–**I**) show the time courses of global solar radiation (Rs) and vapor pressure deficit (VPD). Letters G to I represent the daily values of reference evapotranspiration, $ET_0$ (mm).

Regarding the training systems, the Guyot-trained vines showed higher maximum SF values compared with the Cordon-trained vines over the season (mean of maximum SF values during post-flowering = 0.24 $dm^3$ $h^{-1}$ and 0.16 $dm^3$ $h^{-1}$; post-veraison = 0.18 $dm^3$ $h^{-1}$ and 0.11 $dm^3$ $h^{-1}$; mid-maturation to ripening = 0.18 $dm^3$ $h^{-1}$ and 0.08 $dm^3$ $h^{-1}$, respectively), indicating greater whole-vine transpiration (Figure S2). Taking into account the respective maximum (MXTD) and minimum (MNTD) values, TDFs were mostly similar between training systems during the first interval (e.g., DOY 147: MXTD/MNTD = 1010/866 μm for Guyot vines and 932/783 μm for Cordon vines). Later in the season, these absolute values diminished, though the differences between training systems enlarged (e.g., DOY 220: 759/702 μm and 767/744 μm, respectively), with effects on the CI and MDS results (Section 3.5).

Figure 4 illustrates the daily patterns of SF and TDF on three different days (DOY 147, 199 and 220), each representative of the three phenological intervals defined above for both training systems. On DOY 147 (Figure 4A,D), corresponding to a period of vegetative growth, and in both training systems, SF increased sharply with sunrise, peaked at around 15:00 and decreased to negligible values at the end of the day. On this day, TDF clearly defined a sinusoidal pattern, peaking a maximum value between 6:00 and 8:00, then decreasing between 15:00 and 17:00, followed by a recovery. During the other two sampling days (Figure 4B,C,E,F), TDF flattened its pattern, with similar timing of the maximum value and minimum occurring later (around 18:00–19:00). On the other hand, in these two days, SF reached its maximum earlier (8:00 and 10:00) on Guyot-trained vines (Figure 4E,F) but later on Cordon-trained vines (around 18:00), though a smaller peak was also displayed in the morning (Figure 4B,C).

These daily courses of SF and TDF permit us to define five phases, as described by Herzog et al. [51]. Phase I corresponds to the increase in trunk diameter during the night, when transpiration is considered negligible and the plant rehydrates. Phase II describes the delay between the increase in SF after sunrise and the shrinkage of the trunk. Phase III spans from the beginning of the trunk shrinkage to the maximum SF value (which occurred later). Phase IV corresponds to the delay between the maximum SF and the minimum trunk diameter, normally reached in the afternoon. Finally, phase V describes the delay between the minimum trunk diameter and the minimum SF value. We verified that phases I and II were similar over the three days presented for both training systems (Figure 4). The same did not happen with phases III, IV and V. On DOY 147, the maximum SF was reached after noon (around 14:00), with phase III lasting around 8 h. However, in the remaining two days (DOY 199 and 220), phase III was shorter (about 2–3 h) whereas phase IV was longer compared with DOY 147 for Guyot-trained vines. Regarding Cordon-trained vines, shifts between phases III and IV were displayed on DOY 199 and 220 due to the fact that the maximum SF was reached later than the minimum TDF value (Figure 4B,C). Phase V presented longer durations on DOY 147, with its interval depending on previously phases. The pedoclimatic conditions were mostly responsible for these patterns.

The relationships between daily courses of SF and TDF on the same days presented in Figure 4 are plotted in Figure 5. Lower changes in TDF for the same SF values were found on Cordon-trained vines, where the TDF remained practically unchanged over DOY 199 and 220. Furthermore, in both training systems, trunk diameter fluctuations were clearer in the late afternoon than in the early morning, particularly in DOY 147. For each day, SF responded more sharply than TDF just after sunrise. Rises in SF from early morning would be expected in response to solar radiation (Figure 3G,H,I). However, as was observed previously, FASW and $\Psi_{pd}$ decreased (Figure 2) and VPD tended to increase over maturation (Figure 3H,I), explaining the lower SF values.

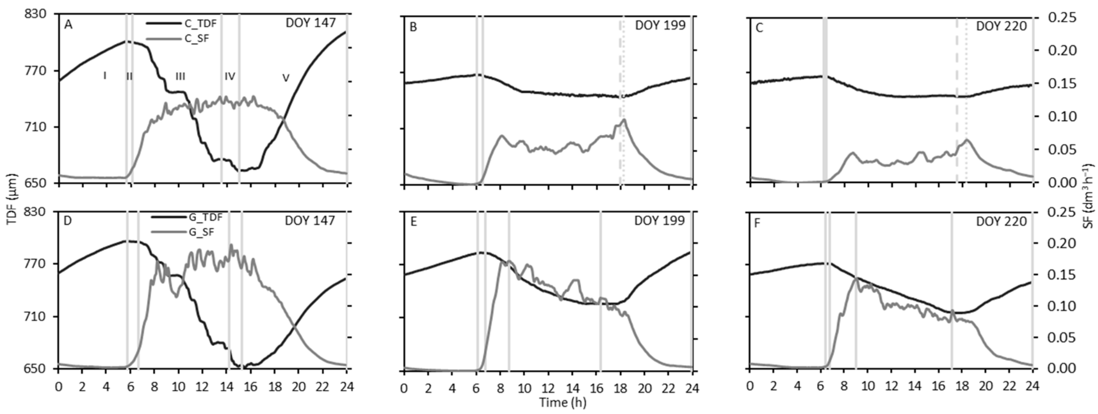

**Figure 4.** Daily courses of sap flow (SF) and trunk diameter fluctuation (TDF) in Touriga-Nacional trained in Cordon (C) system in DOY 147 (**A**), DOY 199 (**B**) and DOY 220 (**C**). Similarly, for Guyot (G) trained vines (**D**–**F**). The five phases described by Herzog et al. [51] are also shown.

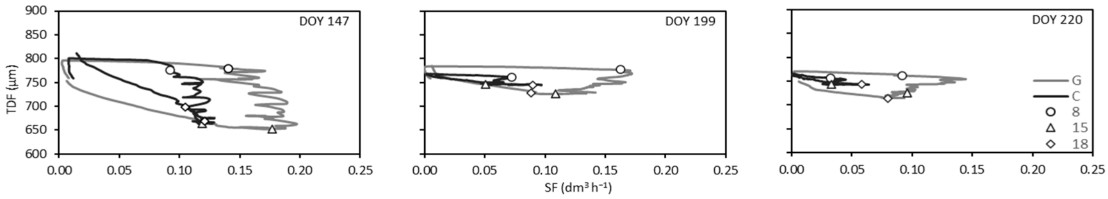

**Figure 5.** Relationship between daily courses of sap flow (SF) and trunk diameter fluctuation (TDF) of Touriga-Nacional trained in Cordon (C) and Guyot (G) systems on three different days of the year (DOY). The values recorded at 8:00, 15:00 and 18:00 are indicated with symbols.

### 3.4. Sap Flow and Leaf Area Index

The leaf area index (LAI) and the ratio between SF and LAI (SF normalized by LAI) are presented in Table 1. LAI increased from DOY 185 to DOY 199 (around veraison), then started to decline over maturation. No significant LAI differences were found between training systems. However, when SF was divided by LAI, the Guyot-trained vines presented significant higher values (mean increase of 38%) compared with Cordon-trained vines, pointing to higher transpiration rates on a whole-plant scale.

**Table 1.** Leaf area index (LAI) and the ratio between sap flow (SF, $dm^3\ d^{-1}$) and LAI in Touriga-Nacional trained in Cordon and Guyot systems on five different days of the year (DOY).

| DOY | | 185 | 199 | 213 | 220 | 234 |
|---|---|---|---|---|---|---|
| **LAI** | Cordon | 0.94 | 1.17 | 0.84 | 0.81 | 0.79 |
| | Guyot | 0.91 | 1.12 | 0.79 | 0.76 | 0.74 |
| *p-value* | | 0.378 | 0.476 | 0.421 | 0.261 | 0.193 |
| **SF/LAI** | Cordon | 1.27 | 1.05 | 1.25 | 1.08 | 0.96 |
| | Guyot | 1.73 | 1.47 | 1.67 | 1.51 | 1.35 |
| *p-value* | | <0.001 | <0.001 | <0.001 | <0.001 | <0.005 |

### 3.5. Seasonal Variation of CI and MDS

Seasonal variation of cumulative increment (CI) and maximum daily trunk shrinkage (MDS) for both training systems are shown in Figure 6. Regarding the Cordon-trained vines, CI increased markedly initially, with a maximum peak at DOY 156 (1682 μm), then decreased sharply to around 1200 μm (DOY 172) and lastly was relatively stable afterwards (Figure 6A). Though the CI pattern was similar for the Guyot-trained vines, they exhibited a higher maximum peak that was reached later (2202 μm on DOY 167). These temporal and absolute differences between training systems were relatively sustained throughout the season (Figure 6A).

The MDS had seasonal behavior similar to the CI, with an initial increase, then decreasing until it became relatively stable, though fluctuations were shown (Figure 6B). Likewise, the Cordon-trained vines presented globally lower MDS values compared with the Guyot-trained vines. The Cordon-trained vines reached their MDS maximums on DOY 143 (173 μm), 146 (165 μm) and 152 (157 μm) and then decreased to around 35 μm. On the other hand, the Guyot-trained vines peaked at a higher MDS maximum (297 μm) and later in the season (DOY 168), also decreasing towards the end of the season (August). MDS had a tendency to increase after irrigation events for both training systems.

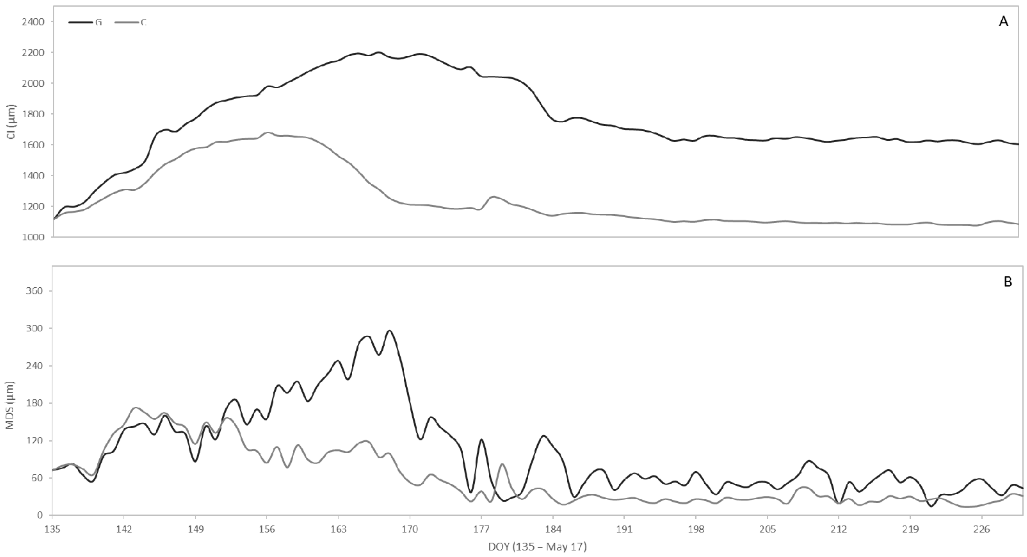


**Figure 6.** Seasonal variation of cumulative increment (CI) (**A**) and maximum daily trunk shrinkage (MDS) (**B**) of Touriga-Nacional trained in Cordon (C) and Guyot (G) systems.

## 4. Discussion

The 2017 growing season stood out, with low precipitation and high temperatures (Figure 1A). The mean temperature for the April–August (April–October) period was 23.4 °C (22.6 °C), clearly above the 30-year average value of 19.0 °C for a similar time period (18.6 °C for April–October; [41]). Annual precipitation was only 55% of the long-term average. A high potential soil water deficit ($ET_0$ minus precipitation) of around 715 mm was calculated from budburst to harvest (Figure 1 and Figure S1). This last value was slightly lower than the 730–750 mm reported by Malheiro [52] for a three-year period in the DDR but with an extended growing season (ca. 20 more days).

Regardless of the water applied by irrigation, a cumulative decrease in FASW was mostly found throughout the season (Figure 2A). This pattern was also reflected in $\Psi_{pd}$ reduction as the summer progressed (Figure 2B), revealing enhanced plant water stress over the season. $\Psi_{pd}$ reflects the equilibrium between soil and leaf water potentials overnight while no significant transpiration is occurring, and it is therefore considered a good indicator of overall plant water status [53]. According to $\Psi_{pd}$ thresholds reported by Ojeda [54] and Van Leeuwen et al. [15], grapevines were exposed to moderate ($\Psi_{pd}$ = −0.45 MPa) to severe ($\Psi_{pd}$ = −0.85 MPa) water deficit during maturation.

During the post-flowering (Figure 3A,D), corresponding to a moderate plant water stress period, daily fluctuations of trunk diameter clearly exhibited three phases: recovery, increment and contraction. The recovery phase corresponds to the cycles during which the trunk radius increases until reaching the maximum value that morning (around 6:00); the increment phase represents the period during which the trunk radius continues to increase until the start of the retraction phase of the next day cycle, and the contraction phase is defined as the period during which the trunk radius decreases, starting from the early morning maximum (starting between 6:00 and 8:00 and ending between 15:00 and 17:00). In the following two periods (Figure 3B,C,E,F), there was a reduction in TDF, with only partial recovery during the night as a reflection of low water availability and high evaporative demand [22,47].

Regarding SF, the curve globally followed the daily course of solar radiation (Figure 3G), with a sharp increase early in the morning (around 6:00; Figure 3A,D) and peaking at high maximum values after noon. When transpiration starts early in the morning, a tension in the evaporative surface of the leaves is created for all plant organs. The water stored in the plant's tissues during the night is then lost, allowing a quick response of the plant to changes in evaporative demand [22,47,55], without depending on the water intake by the roots, which begins later [47]. This explains the rapid increase in SF and the abrupt decrease in TDF during this period (e.g., DOY 147: Figures 4 and 5).

In Figure 3B,C,E,F, a decrease in maximum SF values was evident. This phenomenon was a result of stomatal control in response to lower water availability [24] and higher irradiance and VPD, revealing a key survival strategy of vines under water stress conditions [3,39]. This mechanism has been attributed to abscisic acid (ABA) signaling that induces stomatal closure before a large water deficit develops [56]. Under these atmospheric conditions, irrigation was able to slightly sustain or even increase SF (Figure 3 and Figure S2) but for a short period. In fact, a stomatal closure may also occur even in well-watered vines (which was not the case in the present study) under high atmospheric demand conditions [3]. TDF was also found to be sensitive to water supply in grapevines [55], revealing a small increase in the increment of the trunk but tending to lose these values between irrigation events in our study. Interestingly, daily TDF responded earlier and more visibly than SF during an irrigation event. This finding is in agreement with the results reported by Fernández et al. [35] in olive trees. These last authors also reported a hysteresis phenomenon indicated by clearer trunk diameter fluctuations in the late afternoon than in the early morning. In the present study, similar responses for both training systems were found (Figure 5). On the other hand, Guyot-trained vines showed higher changes in TDF for the same SF values, where TDF of Cordon-vines remained practically unchanged over DOY 199 and 220 (Figure 5). The combined SF and TDF responses allowed us to define the five phases (Figure 4) described by Herzog et al. [51]. Phase III was the most affected from DOY 147 to DOY 199 and 220, being shortened in Guyot-vines and even reversed with phase IV in Cordon-vines. These patterns were related to an earlier sap flow peak (corresponding to earlier stomatal closure) in both training systems and to a later second (and higher) sap flow peak during late afternoon (indicating stomata openness when VPD was decreasing) in Cordon-vines. This response suggests that the Cordon-vines were particularly sensitive to the atmospheric conditions. In fact, stomata openness is related to environmental conditions [4]. On the other hand, under relatively high soil moisture, a reversion between phase I and II was found in olive trees [35], associated with a total recovery of the trunk diameter during the night.

If the soil water is not limiting, transpiration will be conditioned by the leaf area [57]. However, we found that both training systems had similar low/median values of LAI ($0.5 < LAI < 2$; [58]). On the other hand, the SF/LAI ratio was significantly higher for Guyot-trained vines (Table 1), indicating that transpiration per unit area was higher in these vines compared to Cordon-trained vines. In a tall forest species, transpiration per unit leaf area and stomatal conductance per unit leaf area were significantly reduced with increasing crown height [59]. This restriction was a result of the longer length of the hydraulic pathways and enlarged gravitational potential opposing the ascent of water, leading to a reduction in leaf-specific hydraulic conductance [60]. Regarding the grapevines, Favero et al. [61] discussed in their study that the higher trunk height of the Geneva double curtain training system (1.9 m) could have contributed to the difficulty of transporting water to the canopy compared with the 1.0-m vertical shoot positioned trellis. In addition, upward growing of the branch promotes enhanced shoot hydraulic conductivity compared with the downward branch orientation [62]. In this way, our results could be due mainly to the fact that the Guyot-trained vines have lower trunk height and upward growing branches in contrast to the higher trunk and permanent horizontal branch (cordon) of Cordon-trained vines, shortening and facilitating the water pathway along the soil–plant–atmosphere continuum.

During the growing season, CI initially increased and then decreased in both training systems (Figure 6). With decreasing water availability, trunk contraction increases and trunk increment decreases [22,37]. Interestingly, the separation of the fractions of irreversible stem expansion induced by growth and reversible shrinkage and swelling induced by water deficit has been investigated in forest trees in order to better understand the processes of stem radius fluctuations [63,64]. On the other hand, there is evidence that during the fruiting period, the berries become an important sink that can lead to a reduction in the grapevine trunk growth rate [37]. Nonetheless, no significant differences in cluster number and yield were found between training systems in our study (data not shown). Furthermore, according to the last authors, trunk growth ceases regardless of the water status of the plant after veraison. Though LAI reached its maximum value around veraison in the present study (Table 1), CI started decreasing before this phenophase, particularly for Cordon-trained vines (Figure 6A). This response may be associated with the different pedoclimatic conditions between studies.

A similar pattern was found for MDS (Figure 6B). These results are agreement with other studies for grapevines [37] and for other crops, such as olive [27] and apple trees [65]. Furthermore, slight MDS rises were generally found to be due to irrigation (Figure 6B). Conversely, it has been reported that MDS increases under rain-fed conditions compared to an irrigated treatment [37]. The explanation for this apparent discrepancy lies in the occurrence of stress periods in the present study, which prevents the stored tissues from being recharged during the night [47] and promotes a strong reduction in the swelling of the trunk and, therefore, of the MDS [37,47,65]. Guyot-trained vines peaked at higher maximums of CI and MDS values, which were reached later on the season, compared with the Cordon-trained plants. These responses indicate the higher rehydration capacity of the Guyot-trained vines, which may be also associated with the shorter length of its hydraulic pathways [21].

## 5. Conclusions

From a viticultural point of view, the adequate selection of the grapevine training system stands out as one of the most critical adaptation strategies for the present and future times. This study exposed the effect of the shorter length of the hydraulic pathways of the Guyot-trained vines, in contrast to the higher trunk and the permanent horizontal branch (cordon) of the Cordon-trained vines, highlighting the adaptive potential of the Guyot training system to the hot and dry areas. In this way, the results support the selection of the Guyot as a training system that is better adapted to the projected climate change in the DDR as in other Mediterranean wine-growing regions. Stomatal control, in order to reduce transpiration, and rehydration as well as adjust of the total leaf area, were the most important plant survival strategies. The combined use of plant-based measurement techniques, such as sap flow and trunk diameter measurements, revealed sensitivity to variable conditions of atmospheric demand and crop water status. Our results reinforce the usefulness of combining SF and TDF measurements in order to assess grapevine water responses to variable environmental conditions. Further research is needed to comprehend the influence of the hydraulic conductivity of plant segments on grapevine water relations and adaptation measures.

**Supplementary Materials:** The following are available online at http://www.mdpi.com/2077-0472/10/8/315/s1. Figure S1: Monthly mean temperature (line) and total precipitation and reference evapotranspiration, $ET_0$ (bars), recorded at the experimental site from November 2016 to October 2017, Figure S2: Daily means of trunk diameter fluctuations (TDF) and daily values (sums) of sap flow (SF) in Touriga-Nacional trained in the Cordon (C) and Guyot (G) systems during the growing season.

**Author Contributions:** Conceptualization, A.C.M. and J.M.-P.; methodology and investigation, all authors; software, validation, formal analysis and data curation, A.C.M., M.P., N.C. and A.M.C.; resources, A.C.M., L.-T.D. and J.M.-P.; writing—original draft preparation, A.C.M. and M.P.; writing—review and editing, all authors; visualization and supervision, A.C.M.; project administration and funding acquisition, A.C.M. and J.M.-P. All authors have read and agreed to the published version of the manuscript.

**Funding:** This study was funded by the "Grupo Operacional - VITISHIDRI-Estratégias para a gestão do stress hídrico da vinha no Douro Superior", financially supported by the European Agricultural Fund for Rural Development and the Rural Development Programme 2020.

**Acknowledgments:** This work was supported by the European Investment Funds by FEDER/COMPETE/POCI–Operational Competitiveness and Internationalization Programme, under Project POCI-01-0145-FEDER006958, National Funds by FCT—Portuguese Foundation for Science and Technology, under the project UID/AGR/04033/2013. This work was also supported by the project "To Chair–The optimal challenges in irrigation", funded by the FCT. The authors acknowledge Daniel Gomes from "Quinta do Orgal, Vallado" for the collaboration and efforts in making the vineyard facilities available for the study.

**Conflicts of Interest:** The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

## References

1. Van Leeuwen, C.; Friant, P.; Choné, X.; Tregoat, O.; Koundouras, S.; Dubourdieu, D. Influence of climate, soil, and cultivar on terroir. *Am. J. Enol. Vitic.* **2004**, *55*, 207–217.
2. Bernardo, S.; Dinis, L.T.; Machado, N.; Moutinho-Pereira, J. Grapevine abiotic stress assessment and search for sustainable adaptation strategies in Mediterranean-like climates. A review. *Agron. Sustain. Dev.* **2018**, *38*, 66. [CrossRef]
3. Chaves, M.M.; Zarrouk, O.; Francisco, R.; Costa, J.M.; Santos, T.; Regalado, A.P.; Rodrigues, M.L.; Lopes, C.M. Grapevine under deficit irrigation: Hints from physiological and molecular data. *Ann. Bot.* **2010**, *105*, 661–676. [CrossRef] [PubMed]
4. Dinis, L.T.; Correia, C.M.; Ferreira, H.F.; Gonçalves, B.; Gonçalves, I.; Coutinho, J.F.; Ferreira, M.I.; Malheiro, A.C.; Moutinho-Pereira, J. Physiological and biochemical responses of Semillon and Muscat Blanc à Petits Grains winegrapes grown under Mediterranean climate. *Sci. Hortic.* **2014**, *175*, 128–138. [CrossRef]
5. Ferreira, M.I.; Silvestre, J.; Conceição, N.; Malheiro, A.C. Crop and stress coefficients in rainfed and deficit irrigation vineyards using sap flow techniques. *Irrig. Sci.* **2012**, *30*, 433–447. [CrossRef]
6. Fraga, H.; Santos, J.A.; Moutinho-Pereira, J.; Carlos, C.; Silvestre, J.; Eiras-Dias, J.; Mota, T.; Malheiro, A.C. Statistical modelling of grapevine phenology in Portuguese wine regions: Observed trends and climate change projections. *J. Agric. Sci.* **2016**, *154*, 795–811. [CrossRef]
7. Santos, J.A.; Fraga, H.; Malheiro, A.C.; Moutinho-Pereira, J.; Dinis, L.T.; Correia, C.; Moriondo, M.; Leolini, L.; Dibari, C.; Costafreda-Aumedes, S.; et al. A review of the potential climate change impacts and adaptation options for European viticulture. *Appl. Sci.* **2020**, *10*, 3092. [CrossRef]
8. Conejero, W.; Alarcón, J.J.; García-Orellana, Y.; Nicolás, E.; Torrecillas, A. Evaluation of sap flow and trunk diameter sensors for irrigation scheduling in early maturing peach trees. *Tree Physiol.* **2007**, *27*, 1753–1759. [CrossRef]
9. Montoro, A.; Fereres, E.; López-Urrea, R.; Mañas, F.; López-Fuster, P. Sensitivity of trunk diameter fluctuations in *Vitis vinifera* L. Tempranillo and Cabernet Sauvignon cultivars. *Am. J. Enol. Vitic.* **2012**, *63*, 85–93. [CrossRef]
10. Lovisolo, C.; Lavoie-Lamoureux, A.; Tramontini, S.; Ferrandino, A. Grapevine adaptations to water stress: New perspectives about soil/plant interactions. *Theor. Exp. Plant Physiol.* **2016**, *28*, 53–66. [CrossRef]
11. Malheiro, A.C.; Ferreira, M.I.; Conceição, N.; Green, S. Root and stem water dynamics of rainfed grapevines in the Douro region. *Acta Hortic.* **2016**, *1136*, 221–228. [CrossRef]
12. Cifre, J.; Bota, J.; Escalona, J.M.; Medrano, H.; Flexas, J. Physiological tools for irrigation scheduling in grapevine (*Vitis vinifera* L.): An open gate to improve water-use efficiency? *Agric. Ecosyst. Environ.* **2005**, *106*, 159–170. [CrossRef]
13. Simonneau, T.; Lebon, E.; Coupel-Ledru, A.; Marguerit, E.; Rossdeutsch, L.; Ollat, N. Adapting plant material to face water stress in vineyards: Which physiological targets for an optimal control of plant water status? *OENO One* **2017**, *51*, 167–179. [CrossRef]
14. Smart, R.E. Principles of Grapevine Canopy Microclimate Manipulation with Implications for Yield and Quality. A Review. *Am. J. Enol. Vitic.* **1985**, *36*, 230–239.

15. Van Leeuwen, C.; Tregoat, O.; Choné, X.; Bois, B.; Pernet, D.; Gaudillére, J.P. Vine water status is a key factor in grape ripening and vintage quality for red bordeaux wine. How can it be assessed for vineyard management purposes? *J. Int. Sci. Vigne Vin* **2009**, *43*, 121–134. [CrossRef]
16. Reynolds, A.G.; Vanden Heuvel, J.E. Influence of grapevine training systems on vine growth and fruit composition: A review. *Am. J. Enol. Vitic.* **2009**, *60*, 251–268.
17. Fragasso, M.; Antonacci, D.; Pati, S.; Tufariello, M.; Baiano, A.; Forleo, L.R.; Caputo, A.R.; La Notte, E. Influence of training system on volatile and sensory profiles of primitivo grapes and wines. *Am. J. Enol. Vitic.* **2012**, *63*, 477–486. [CrossRef]
18. Ferree, D.; Steiner, T.; Gallander, J.; Scurlock, D.; Johns, G.; Riesen, R. Performance of "Seyval Blanc" grape in four training systems over five years. *HortScience* **2002**, *37*, 1023–1027. [CrossRef]
19. Cavallo, P.; Poni, S.; Rotundo, A. Ecophysiology and vine performance of cv. "Aglianico" under various training systems. *Sci. Hortic.* **2001**, *87*, 21–32. [CrossRef]
20. IVV. *Vinhos e Aguardentes de Portugal, Anuário 2018*; Ministério da Agricultura, do Desenvolvimento Rural e das Pescas, Instituto da Vinha e do Vinho: Lisboa, Portugal, 2018.
21. Magalhães, N. *Tratado de Viticultura: A Videira, a Vinha e o "Terroir"*, 1st ed.; Esfera Poética: Lisboa, Portugal, 2015; 608p, ISBN 978-972-8987-15-2.
22. Fernández, J.E. Plant-based methods for irrigation scheduling of woody crops. *Horticulturae* **2017**, *3*, 35. [CrossRef]
23. Meng, Z.; Duan, A.; Chen, D.; Dassanayake, K.B.; Wang, X.; Liu, Z.; Liu, H.; Gao, S. Suitable indicators using stem diameter variation-derived indices to monitor the water status of greenhouse tomato plants. *PLoS ONE* **2017**, *12*, e0171423. [CrossRef] [PubMed]
24. Escalona, J.; Flexas, J.; Medrano, H. Drought effects on water flow, photosynthesis and growth of potted grapevines. *Vitis* **2002**, *41*, 57–62.
25. Daudet, F.A.; Améglio, T.; Cochard, H.; Archilla, O.; Lacointe, A. Experimental analysis of the role of water and carbon in tree stem diameter variations. *J. Exp. Bot.* **2005**, *56*, 135–144. [CrossRef] [PubMed]
26. Velez, J.E.; Intrigliolo, D.S.; Castel, J.R. Scheduling deficit irrigation of citrus trees with maximum daily trunk shrinkage. *Agric. Water Manag.* **2007**, *90*, 197–204. [CrossRef]
27. Moriana, A.; Girón, I.F.; Martín-Palomo, M.J.; Conejero, W.; Ortuño, M.F.; Torrecillas, A.; Moreno, F. New approach for olive trees irrigation scheduling using trunk diameter sensors. *Agric. Water Manag.* **2010**, *97*, 1822–1828. [CrossRef]
28. de la Rosa, J.M.; Conesa, M.R.; Domingo, R.; Torres, R.; Pérez-Pastor, A. Feasibility of using trunk diameter fluctuation and stem water potential reference lines for irrigation scheduling of early nectarine trees. *Agric. Water Manag.* **2013**, *126*, 133–141. [CrossRef]
29. Granier, A. Une nouvelle méthode pour la mesure du flux de sève brute dans le tronc des arbres. *Ann. Sci. For.* **1985**, *42*, 193–200. [CrossRef]
30. de Swaef, T.; Steppe, K. Linking stem diameter variations to sap flow, turgor and water potential in tomato. *Funct. Plant Biol.* **2010**, *37*, 429–438. [CrossRef]
31. Ortuño, M.F.; García-Orellana, Y.; Conejero, W.; Ruiz-Sánchez, M.C.; Mounzer, O.; Alarcón, J.J.; Torrecillas, A. Relationships between climatic variables and sap flow, stem water potential and maximum daily trunk shrinkage in lemon trees. *Plant Soil* **2006**, *279*, 229–242. [CrossRef]
32. Ortuño, M.F.; García-Orellana, Y.; Conejero, W.; Ruiz-Sánchez, M.C.; Alarcón, J.J.; Torrecillas, A. Stem and leaf water potentials, gas exchange, sap flow, and trunk diameter fluctuations for detecting water stress in lemon trees. *Trees Struct. Funct.* **2006**, *20*, 1–8. [CrossRef]
33. Coehn, M.; Goldhamer, D.A.; Fereres, E.; Girona, J.; Mata, M. Assessment of peach tree responses to irrigation water ficits by continuous monitoring of trunk diameter changes. *J. Hortic. Sci. Biotechnol.* **2001**, *76*, 55–60. [CrossRef]
34. Conejero, W.; Alarcón, J.J.; García-Orellana, Y.; Abrisqueta, J.M.; Torrecillas, A. Daily sap flow and maximum daily trunk shrinkage measurements for diagnosing water stress in early maturing peach trees during the post-harvest period. *Tree Physiol.* **2007**, *27*, 81–88. [CrossRef] [PubMed]
35. Fernández, J.E.; Moreno, F.; Martín-Palomo, M.J.; Cuevas, M.V.; Torres-Ruiz, J.M.; Moriana, A. Combining sap flow and trunk diameter measurements to assess water needs in mature olive orchards. *Environ. Exp. Bot.* **2011**, *72*, 330–338. [CrossRef]

36. Cuevas, M.V.; Torres-Ruiz, J.M.; Álvarez, R.; Jiménez, M.D.; Cuerva, J.; Fernández, J.E. Assessment of trunk diameter variation derived indices as water stress indicators in mature olive trees. *Agric. Water Manag.* **2010**, *97*, 1293–1302. [CrossRef]
37. Intrigliolo, D.S.; Castel, J.R. Evaluation of grapevine water status from trunk diameter variations. *Irrig. Sci.* **2007**, *26*, 49–59. [CrossRef]
38. Baert, A.; Villez, K.; Steppe, K. Automatic drought stress detection in grapevines without using conventional threshold values. *Plant Soil* **2013**, *369*, 439–452. [CrossRef]
39. Oliveira, M.T. Dependence of Sap Flow and Stem Diameter Fluctuation of Grapevines on Reference Evapotranspiration: An Event Coincidence Analysis. *Adv. Plants Agric. Res.* **2017**, *7*, 433–439. [CrossRef]
40. Kottek, M.; Grieser, J.; Beck, C.; Rudolf, B.; Rubel, F. World map of the Köppen-Geiger climate classification update. *Meteorol. Z.* **2006**, *15*, 259–263. [CrossRef]
41. Fonseca, A.R.; Santos, J.A. High-resolution temperature datasets in Portugal from a geostatistical approach: Variability and extremes. *J. Appl. Meteorol. Climatol.* **2018**, *57*, 627–644. [CrossRef]
42. Allen, R.G.; Pereira, L.S.; Raes, D.; Smith, M. *Crop Evapotranspiration: Guidelines for Computing Crop Water Requirements*; FAO Irrigation and Drainage Paper 56; Food and Agriculture Organization of the United Nations: Rome, Italy, 1998; ISBN 9251042195.
43. Agroconsultores and Coba. *Carta de Solos, Carta de Uso Actual da Terra e Carta de Aptidão da Terra, do Nordeste de Portugal*; Universidade de Trás-os-Montes e Alto Douro (UTAD): Vila Real, Portugal, 1991.
44. Granier, A.; Gross, P. Mesure du flux de sève brute dans le tronc du Douglas par une nouvelle méthode thermique. *Ann. Sci. For.* **1987**, *44*, 1–14. [CrossRef]
45. Katerji, N.; Tardieu, F.; Bethenod, O.; Quetin, P. Behavior of Maize Stem Diameter during Drying Cycles: Comparison of Two Methods for Detecting Water Stress. *Crop Sci.* **1994**, *34*, 165–169. [CrossRef]
46. Zweifel, R.; Item, H.; Häsler, R. Stem radius changes and their relation to stored water in stems of young Norway spruce trees. *Trees Struct. Funct.* **2000**, *15*, 50–57. [CrossRef]
47. Fernández, J.E.; Cuevas, M.V. Irrigation scheduling from stem diameter variations: A review. *Agric. For. Meteorol.* **2010**, *150*, 135–151. [CrossRef]
48. Scholander, P.F.; Hammel, H.J.; Bradstreet, A.; Hemmingsen, E.A. Sap Pressure in Vascular Plants. *Science* **1965**, *148*, 339–346. [CrossRef] [PubMed]
49. Turner, N.C. Measurement of plant water status by the pressure chamber technique. *Irrig. Sci.* **1988**, *9*, 289–308. [CrossRef]
50. López-Lozano, R.; Casterad, M.A. Comparison of different protocols for indirect measurement of leaf area index with ceptometers in vertically trained vineyards. *Aust. J. Grape Wine Res.* **2013**, *19*, 116–122. [CrossRef]
51. Herzog, K.M.; Häsler, R.; Thum, R. Diurnal changes in the radius of a subalpine Norway spruce stem: Their relation to the sap flow and their use to estimate transpiration. *Trees* **1995**, *10*, 94–101. [CrossRef]
52. Malheiro, A.C. Microclimate, Yield and Water-Use of Vineyards in the Douro Region, Portugal. Ph.D. Thesis, Cranfield University, Bedfordshire, UK, 2005.
53. Winkel, T.; Rambal, S. Influence of Water Stress on Grapevines Growing in the Field: From Leaf to Whole-Plant Response. *Aust. J. Plant Physiol.* **1993**, *20*, 143–157. [CrossRef]
54. Ojeda, H.; Saurin, N. L'irrigation de précision de la vigne: Méthodes, outils et stratégies pour maximiser la qualité et les rendements de la vendange en économisant de l'eau. *Innov. Agron.* **2014**, *38*, 97–108.
55. Santesteban, L.G.; Palacios, I.; Miranda, C.; Iriarte, J.C.; Royo, J.B.; Gonzalo, R. Terahertz time domain spectroscopy allows contactless monitoring of grapevine water status. *Front. Plant Sci.* **2015**, *6*, 404. [CrossRef]
56. Kramer, P.J. *Water Relations Plants*; Academic Press: New York, NY, USA, 1983.
57. Pereira, A.R.; Green, S.; Villa Nova, N.A. Penman-Monteith reference evapotranspiration adapted to estimate irrigated tree transpiration. *Agric. Water Manag.* **2006**, *83*, 153–161. [CrossRef]
58. Pearce, I.; Coombe, B.G. Grapevine phenology. In *Viticulture*; Dry, P.R., Coombe, B.G., Eds.; Winetitles: Broadview, Australia, 2005.
59. Ambrose, A.R.; Sillett, S.C.; Koch, G.W.; Van Pelt, R.; Antoine, M.E.; Dawson, T.E. Effects of height on treetop transpiration and stomatal conductance in coast redwood (*Sequoia sempervirens*). *Tree Physiol.* **2010**, *30*, 1260–1272. [CrossRef] [PubMed]
60. Ryan, M.G.; Phillips, N.; Bond, B.J. The hydraulic limitation hypothesis revisited. *Plant Cell Environ.* **2006**, *29*, 367–381. [CrossRef] [PubMed]

61. Favero, A.C.; Amorim, D.A.D.; Mota, R.V.D.; Souza, C.R.D.; Regina, M.D.A. Physiological responses and production of "Syrah" vines as a function of training systems. *Sci. Agric.* **2010**, *67*, 267–273. [CrossRef]
62. Schubert, A.; Lovisolo, C.; Peterlunger, E. Shoot orientation affects vessel size, shoot hydraulic conductivity and shoot growth rate in *Vitis vinifera* L. *Plant Cell Environ.* **1999**, *22*, 197–204. [CrossRef]
63. Zweifel, R. Radial stem variations—A source of tree physiological information not fully exploited yet. *Plant Cell Environ.* **2016**, *39*, 231–232. [CrossRef]
64. Zweifel, R.; Haeni, M.; Buchmann, N.; Eugster, W. Are trees able to grow in periods of stem shrinkage? *New Phytol.* **2016**, *211*, 839–849. [CrossRef]
65. Liu, C.; Kang, S.; Li, F.; Li, S.; Du, T.; Tong, L. Relationship between environmental factor and maximum daily stem shrinkage in apple tree in arid region of northwest China. *Sci. Hortic.* **2011**, *130*, 118–125. [CrossRef]